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## A STRUCTURAL FEATURE CONNECTED WITH THE MATING OF DIEMYCTYLUS VIRIDESCENS.

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THERE seems to be very little doubt at present that the fertilization of Urodela is internal. Since Spallanzi in 1785 proved internal fertilization must take place with Triton, many investigators have shown conclusively that such is the case in other species.

The mating habits of a number of salamanders have been observed, but very much is yet to be learned about many forms concerning which we now know little more than the mere fact that fertilization is internal; however, it seems very probable that in all Urodela fertilization is accomplished by a similar method. In most cases the courtship is more or less complicated and terminates with the female following the male, who emits a spermatophore containing zoösperms, which is received by the cloaca of the female as she passes over it.

There are many variations in the mating habits of different forms described, but unfortunately few species are easy to observe. Of American forms practically nothing is known of the more terrestrial species. *Diemyctylus viridescens* and *D. torosus* of the aquatic species are about the only ones in which a satisfactory idea of the mating has been obtained.

A number of observations have been made upon European salamanders by Robin ('74), Gasco ('80), and Zeller ('90), who have given good accounts of their habits. With Triton, for instance, Zeller has carefully described the antics of the male before the female; these may extend over a period of hours; he rubs against her and rushes before her. At length she is attracted by him, and as he moves off she follows and receives

in her cloaca the spermatophore which he emits. This method of mating is very commonly described among salamanders, but variations occur; for instance, in *Salamandra maculosa*, and in *Salamandra atra*, Siebold ('58), Pfitzner ('80), and others describe the male perched upon the female's back; he clasps her for a period before spermatophores are voided. In these cases the male is usually perched upon the center of the female's back. In *Megapterna montana* and several others Bedriaga ('82) says the male grasps the female and emits spermatophores while still clasping her.

Ritter ('99) in *D. torosus* described, so far as he was able, a method of fertilization similar to that given by Bedriaga in several species; that is, the male emits spermatozoa while clasping the female. The mating of *D. viridescens* has been carefully described by Zeller ('90), Gage ('91), and Jordan ('91), and, so far as the final result is concerned, the process may be said to be similar to that described by Zeller and others for many forms of Salamandridæ, but in several particulars the mating habits are peculiar and deserve further attention.

As breeding season approaches with *Diemyctylus*, a crest appears upon the already broad tail of the male, and the cloacal regions of both male and female enlarge. It is stated by some that during this time the colors of the male become brighter; but this is probably exaggerated, as there is but little difference in the color of the male and female at this time, except possibly one or the other may have recently passed the red land stage and not yet attained the deeper color of the more mature individuals, or it may have just shed its skin. With *Diemyctylus* there is much less difference in the sexes than is found in many European Urodela, yet there are several striking differences, most of which have been noted. The female differs from the male at all seasons of the year by having more slender hind legs, and during breeding season there are on the undersides of the male's hind legs black, wartlike, horny elevations, probably developed to aid the male in clasping the female. These have often been described, but as this method of clasping the female is rare with European species, the occurrence of these organs is seldom mentioned with them.

Cope ('89) in his work on Batrachia describes a series of three to four pits along the sides of the head just back of the eye; he regards these as not constant for the species (Fig. 1).

After examining a considerable number of animals I have come to the conclusion that these pits are constant structures. In adult males there are usually three to four quite large, deep pits, which can be seen without difficulty extending from just back of the eye along the side of the head. In adult females, as a general thing, one would say there are no pits, but on a more careful examination one to four small depressions are often seen; these usually appear as pin pricks, or minute depressions in the skin. They are similar in structure to those found in the adult males, but are much smaller and less developed in the female.

In early larval forms there are no signs of depressions or pits in either males or females, but in medium-sized to large red males there are along the sides of the head minute depressions which look very much like those found in the adult females, and more or less advanced, according to the size of the animal.

In early red females, as a usual thing, no pits are found, but in later red forms small beginning pits are sometimes present and sometimes absent; but judging from all the individuals examined it may be stated that as a usual thing these small pits of the female make their appearance at about the time the red land form changes to the viridescent water form, and that the corresponding structures in the male make their appearance in the small red forms about the time that their sex can be determined by dissection.

Before speaking of the significance of these structures which are so well developed in the adult male and so rudimentary in the female and young, it will be necessary to speak somewhat further of the mating habits. Usually in captivity during the fall, winter, and spring the male at once settles upon the female's back and clasps her just in front of her fore legs with his powerful back legs in such a manner that it is impossible for her to escape and, at the same time, possible for him to

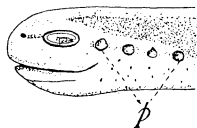


FIG. 1. — Side view of the head of an adult male *Diemyctylus*. *p*, pits. About natural size.

bring his head down in contact with hers. Although she is unable to escape, in many cases she may struggle, carrying the male with her for a short distance ; but usually it is the other way,

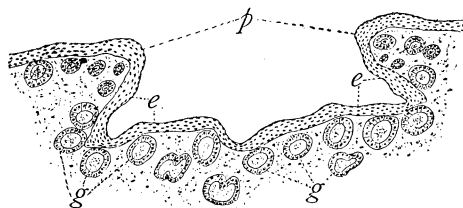


FIG. 2. — Section of a pit and glands of an adult male *Diemyctylus*. *g*, gland tubules; *e*, epithelium of pit.  $\times 30$ .

and the male jerks the female about after having been perched quietly on her back for a short time. This jerking about is not without system, for almost invariably the male jerks the female somewhat and places one side of his head just below the top and a little back of the eye, in contact with that of the female, leaving his head there for a few seconds and waving his tail with a gentle fanning motion. After a short period he jerks the female about in such a way that the other side of his head occupies a similar position in regard to the female's nose, pulling her violently in order to do this. This touching one side of his head and then the other side to the female's snout, accompanied by the more or less necessary jerking about in the water and fanning movements of the tail, continue for some time, and after a considerable number of these jerkings the male slowly leaves the female, his cloaca expanded, and is usually followed closely by the female, whose nose is near his tail. The male moves slowly forward and throws his body into serpentine undulations, and a spermatophore is emitted which may come to the cloaca of the female as she follows him farther along.

The question naturally arises as to what relation these constant habits have towards the accomplishment of fertilization; of course it is evident that it is necessary for the male to be followed by the female in order that she may receive the spermatozoa which he emits in masses or spermatophores, and her following him may be due to an attraction which he exerts over her, but what is the nature of this attraction ?

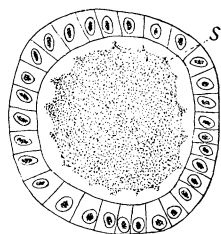


FIG. 3. — Section of a tubule of an adult male *Diemyctylus*. *s*, secretion.  $\times 210$ .

As indicated by Jordan ('91), and as simple experiments in feeding captive *Diemyctylus* show, probably the strongest sense of the newt is either the sense of smell or the tactile sense; the sense of sight is not particularly well developed. Now, to return to the pits on either side of the head; upon sectioning these pits it was found that a number of simple gland tubes were collected about the bases of the pits, and these tubules (Figs. 2, 3, 4), although not opening widely in the pits when not active, may be seen to have places where the cell nuclei were drawn out and arranged parallel with the epithelial nuclei; but when the glands are filled with secretion a few well-marked openings into the pits may be found (Fig. 5). These pits are formed from simple depressions or ingrowths of epithelium, and the epithelium which lines the pits does not differ in any way from that which covers the surface of the body (Fig. 6). The pits were found to begin in about half-grown red males or very late red females. As adult life is reached these pits or depressions become broader, and usually deeper, and together with

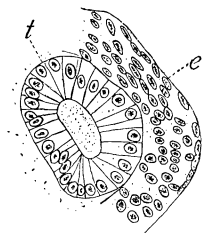


FIG. 4. — Section of a tubule and pit epithelium from an inactive gland of an adult male *Diemyctylus*. *t*, tubule; *e*, epithelium.  $\times 210$ .

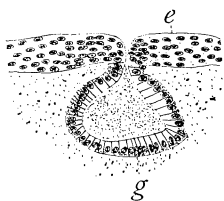


FIG. 5. — Section of a tubule from an adult male *Diemyctylus*, showing the opening into a pit. *e*, epithelium; *g*, gland tubule.  $\times 165$ .

this broadening and deepening there is an increased development of the glands, which become very numerous in adult males.

The glands are first formed when the pits are hardly more than narrow insinkings of the skin, and, as Ancel (1901) states for other skin glands of Amphibia, they are derived from the ectoderm. The cells which form the rudiments of these glands are derived from those of the insinking, or directly from the adjoining surfaces of the skin (Fig. 7). The little masses of cells which develop into glands become nearly separated from the epithelium

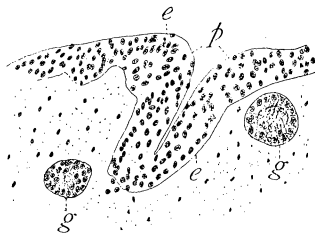


FIG. 6. — Section of a pit in a young red male *Diemyctylus*. *e*, epithelium; *g*, rudimentary gland tubules.  $\times 105$ .

from which they were derived, and the communications with the exterior by means of minute ducts are not formed until much later. The cells of the early rudiments of glands soon begin to be arranged in a more and more columnar manner, and after they are arranged in this way the duct to the exterior is formed partly from cells of the gland itself and

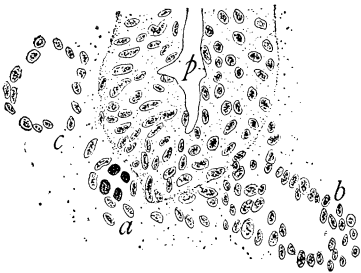


FIG. 7. — Section of a pit of a young red male *Diemictylus*. *p*, pit; *a*, early stage of a developing gland; *b*, later stage of developing gland; *c*, still later stage of developing gland.  $\times 210$ .

partly from the epithelium of the pit. The fully developed gland is simple, saccular, or slightly tubular, with a very small duct. Sometimes glands are found which seem to be partially divided into several portions; that is, have one, two, or more lumens branching off from the main one (Figs. 8, 9). New glands are formed from the old ones in this way, and although these dividing tubules are found most abundantly in half-grown specimens, a few of them are often found in the adult. These glands are more perfectly formed in those individuals where the pits are best developed; but those in a very active condition, with their lumens widely distended, were only found in adult males during the breeding season. During the mating season, either in fall or spring, these glands were very much enlarged in the adult males, their epithelial cells were much like low cubes, and the tubules possessed large lumens (Figs. 3, 10).

When these glands were sectioned out of breeding season a very marked contrast was noticed; their lumens were much diminished, the cells reached and nearly touched in the center and no colloid secretion was present (Fig. 4). These little pits of the immature animals or of the adult females have at their bases a few masses of cells, or a few small gland tubules, which are undoubtedly the beginnings or remnants of the

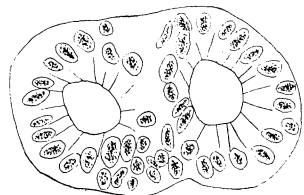


FIG. 8. — Section of a tubule from a red male *Diemictylus* of nearly adult size.  $\times 260$ .

glands so characteristic of the adult males (Fig. 11); sometimes one or two tubules appear to have secretion in the lumens, and some have openings into the pits.

To recapitulate: The pits in the adult males are almost invariably well marked, while those in the females, when they occur, are rather difficult to detect; and then, too, the gland tubules in the male are very numerous (Fig. 2).

Adult males taken at other times than the breeding season, as a usual thing, show the gland tubules in a resting state; the lumens of the glands commonly have the cells almost touch-

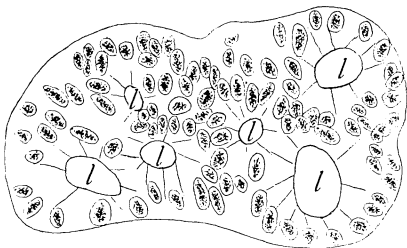


FIG. 9. — Section of a tubule from a red male *Diemyctylus* of nearly adult size. L, lumen.  $\times 260$ .

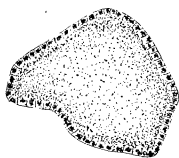


FIG. 10. — Section of a gland tubule from an adult male; taken during mating. The tubule is filled with secretion.  $\times 105$ .

ing each other at the center, but males taken when breeding show these gland lumens filled with secretion and the cells appear as a low epithelium.

It may be that these glands which are developed so extensively in the male, and placed in a position which favors them, are for the purpose of attracting the female by means of some secretion.

Although, as already spoken of, similar mating habits have in general been described, they are not exactly the same as in *D. viridescens*, nor have similar glands or pits been described for other forms. Ritter, in an interesting paper on *Diemyctylus torosus*, describes no such structures, nor was I able to detect any trace of such pits or glands in several specimens examined; and, in fact, as already stated, the mating habits are entirely dissimilar in the two species.

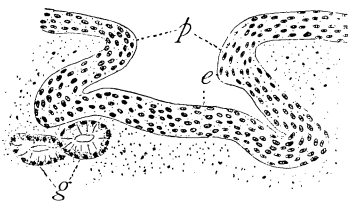


FIG. 11. — Section of a pit from an adult female *Diemyctylus*. e, epithelium of pit; g, rudimentary gland.  $\times 105$ .



## GENERAL SUMMARY.

1. The glands on the side of the head of *D. viridescens* reach their complete development only in adult males, and according to my observations are most active during the time of mating.

2. The pits are formed from invaginations of the epidermis, and the gland rudiments are derived from these invaginations or from the adjoining epidermal surfaces.

3. Adult males always possess three to four large, well-marked pits. Adult females sometimes have one to four pits on a side, and also a few small gland tubules, some of which may open into the pits.

4. Early red larval males have small pits about as soon as the sex can be determined by gross dissection, and at the bases of the pits there are usually a few small gland tubules.

5. Pits when present in red females make their appearance *late*, usually just before the red terrestrial changes to the viridescent aquatic form; at this time the pits are seen to be shallow depressions of the skin, and no gland tubules are present.

6. No pits or glands were found in the early larval form of *D. viridescens*.

7. The glands described occur only when pits are present, and the tubules have openings into the pits in the adult males, and sometimes in the females.

8. In mating, the female follows the male only after the usual courtship, in which first one side and then the other side of the male's head touches the female's snout.

In the preparation of this paper I have become greatly indebted to the Department of Histology of Cornell University and the professors in charge, and also to Mrs. Gage for lending me her slides of *Diemyctylus* heads to complete my series of stages.

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